

(19)



Europäisches Patentamt

European Patent Office

Office européen des brevets



(11)

EP 0 593 774 B1

(12)

EUROPEAN PATENT SPECIFICATION

(45) Date of publication and mention
of the grant of the patent:
06.08.1997 Bulletin 1997/32

(51) Int. Cl.⁶: **A23L 1/30, A23L 2/44,**
A23L 2/40, A23G 3/00,
A23L 1/09, A23L 1/236

(21) Application number: 93906886.2

(86) International application number:
PCT/JP93/00436

(22) Date of filing: 02.04.1993

(87) International publication number:
WO 93/20718 (28.10.1993 Gazette 1993/26)

**(54) FOOD COMPOSITION WHICH INHIBITS FORMATION OF INTESTINAL PUTREFACTION
PRODUCT**

NAHRUNGSMITTELZUSAMMENSETZUNG ZUR HEMMUNG VON DER BILDUNG VON
DARMVERROTTUNGSPRODUKTEN

COMPOSITION ALIMENTAIRE INHIBANT LA FORMATION DE PRODUITS DE PUTREFACTION
INTESTINALE

(84) Designated Contracting States:
CH DE ES FR GB IT LI NL

• SAKAMOTO, Shuichi,
Room No. 304 Daiyaparesu Suwano
Kurume-shi, Fukuoka 830 (JP)

(30) Priority: 10.04.1992 JP 91185/92

• OKAMATSU, Hiroshi
Kurume-chi, Fukuoka 830 (JP)

(43) Date of publication of application:
27.04.1994 Bulletin 1994/17

• KUMEMURA, Megumi,
Room No. 608,
Kurume-chi, Fukuoka 830 (JP)

(73) Proprietor:
OTSUKA PHARMACEUTICAL CO., LTD.
Chiyoda-ku Tokyo 101 (JP)

(74) Representative: Hansen, Bernd, Dr. Dipl.-Chem.
et al
Hoffmann Eitle,
Patent- und Rechtsanwälte,
Postfach 81 04 20
81904 München (DE)

(72) Inventors:

- TAKAICHI, Akihisa
Naruto-shi, Tokushima 772 (JP)
- OKAMOTO, Toshihiko
Tokushima-shi, Tokushima 779-31 (JP)
- AZUMA, Yoshihide,
Room No. 701, Mezon Kitahama
Muya-cho, Naruto-shi, Tokushima 772 (JP)
- WATANABE, Yoshinari
Itano-gun, Tokushima 771-12 (JP)
- MATSUMOTO, Toshiaki,
Room No. 101, Kopo Mori
Tokushima-shi, Tokushima 770 (JP)
- MIYATA, Katsuya,
Otsuka Imagire-ryo
Tokushima-shi, Tokushima 771-01 (JP)

(56) References cited:
EP-A- 0 447 125 EP-A- 0 587 904
JP-A- 4 099 472 JP-A-52 083 731
JP-A-54 044 013 JP-A-62 220 169
JP-B-49 004 377

- Japan Food Science, Vol. 30, No. 12, (1991),
KOZO HARA, "Characteristics of dairy
oligosaccharide (lactoseclos) and its application
to processed foods", p. 46-61.
- New Food Industry, Vol. 33, No. 9, (1991),
YOSHIHIDE OZAKI, "Characteristics of dairy
oligosaccharide (lactoseclos) and its application
- bifidus proliferation sugar", p. 12-15.

Note: Within nine months from the publication of the mention of the grant of the European patent, any person may give notice to the European Patent Office of opposition to the European patent granted. Notice of opposition shall be filed in a written reasoned statement. It shall not be deemed to have been filed until the opposition fee has been paid. (Art. 99(1) European Patent Convention).

Description

The present invention relates to a novel drink composition for inhibiting the formation of an intestinal putrefactive product.

It is known that skatole, indole, p-cresol, 4-ethylphenol and the like are intestinal putrefactive product derived from tryptophan, tyrosine and the like, and may be promoters of a variety of cancers. It is therefore desired to inhibit such components from being formed in intestines.

EP-A-0 447 125 discloses a food product for inhibiting the formation of an intestinal putrefactive product, comprising lacto-sucrose.

It is a main object of the present invention to provide a drink composition for inhibiting the formation of an intestinal putrefactive product, which can reduce the amount of a harmful putrefactive product to be formed in intestines.

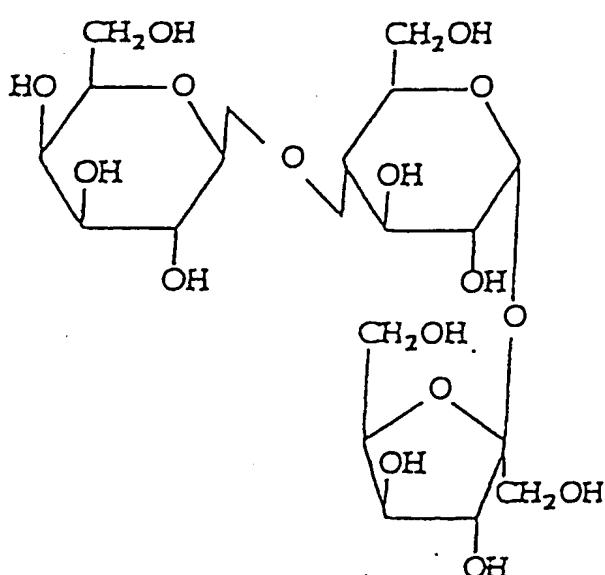
It is another object of the present invention to provide a drink composition for inhibiting the formation of an intestinal putrefactive product, which can inhibit an effective component from being decomposed to enhance the stability of the effective component.

After hard study for achieving the objects above-mentioned, the inventors have found the following surprising fact, based on which the present invention has been completed. That is, the intake of lactosucrose can not only reduce the amount of a putrefactive product such as p-cresol, skatole, indole or the like to be generated in human stool, but also lowers the rate of detection, in stool, of germs which contribute to the formation of such a putrefactive product.

The invention is directed to a drink composition for inhibiting the formation of an intestinal putrefactive product, containing lactosucrose in an amount of 0.5 to 30 g/100 ml and a buffer solution being added to said composition such that the pH of said composition is adjusted in the range from 4.0 to 6.5.

Lactosucrose is oligosaccharide and can accelerate the propagation of intestinal bifid bacteria, thus reducing the amount of the putrefactive product above-mentioned to be formed. Further, lactosucrose is indigestible and therefore very low in calorific value. Accordingly, lactosucrose is suitable for low-caloric food. The drink composition of the present invention containing lactosucrose as an effective component, is fully satisfactory in view of taste, odor, dietary feeling and the like.

Lactosucrose used in the present invention is O - β - D - galactopyranosyl - (1 \rightarrow 4) - O - α - D - glucopyranosyl - (1 \leftarrow 2) - β - D - fructofuranoside, represented by the following formula:



There may be used such a substance which is produced by a conventional producing method. Examples of the conventional producing method include (i) a method discussed in Japanese Patent Publication No. 57-58905 in which, for example, levan sucrase originating from genus Aerobacter, is acted on a solution of sucrose and lactose, (ii) a method discussed in Japanese Patent Unexamined Publication No. 64-85090 in which an extract of cells from the specific genus sporobolomyces, are used, and (iii) a method discussed in Japanese Patent Unexamined Publication No. 2-35095 in which germs of the genus Rohnella are used. In the present invention, lactosucrose produced by any of these methods may be used as it is, or as refined by column chromatography.

The drink composition of the present invention is not limited in form, but may be used in the form of a liquid, a syrup, or a powder. Further, the drink composition of the present invention may suitably contain extenders, sweetners, vitamins, cells of bifid bacteria and the like. From the drink composition of the present invention, there may be formed (i) a liquid or powdery sweetner such as sirup, (ii) a drink such as a refreshing drink, a milk beverage or the like,

5 In the drink composition of the present invention, lactosucrose is contained in an amount of 0.5 to 30 g, preferably 1 to 15 g, for 100 ml of such a drink.

In a drink containing lactosucrose, to restrain pH from being lowered to stably maintain the lactosucrose for a long period of time, the present invention uses a buffer solution to maintain pH in the range from 4.0 to 6.5, preferably from 4.5 to 6.0. If pH is too low, lactosucrose is liable to be decomposed. On the other hand, as pH is increased, the drink is liable to lack an organic refreshing feeling. It is therefore preferable to set pH in the range above-mentioned. When carbonic acid is added, a refreshing feeling is increased and sterilizing conditions such as heating temperature and time can be relaxed, thus further improving the stability of lactosucrose.

10 As the buffer agent, there may be used a mixture solution containing a weak acid having a buffer function and its salt. The weak acid and its salt may be blended with a drink composition such that the drink composition presents the target pH.

15 Examples of the weak acid include citric acid, tartaric acid, lactic acid, malic acid, carbonic acid and the like. Examples of the weak acid salt include sodium citrate, sodium tartrate, sodium malate, calcium lactate, sodium lactate, sodium hydrogenphosphate, sodium carbonate, sodium hydrogencarbonate and the like.

20 A buffer agent comprising a weak acid and its salt, may be blended in such a necessary amount as to maintain pH of the drink composition in the range above-mentioned. That is, the blending amount of the buffer agent is suitably determined according to the type of a drink composition, but may be in the range preferably from 0.03 to 2 % by weight and more preferably from about 0.05 to about 0.3 % by weight.

25 In the drink composition of the present invention which contains lactosucrose as an effective component, any of a variety of glucides and sweetners may be added as done in normal drinks. Examples of the glucide component include (i) a variety of saccharides including a monosaccharide such as glucose, fructose and the like, and a disaccharide such as maltose, sucrose and the like, (ii) polysaccharide such as dextrin, cyclodextrin and the like, and (iii) sugar alcohols such as xylitol, erythritol, sorbitol and the like. Examples of the sweetening agent include natural sweetners (thaumatin, an extract of stevia, a glycyrrhizin), and synthetic sweetners (saccharin, aspartame and the like). These glucide components and sweetners may be blended in an amount of normally 15 % by weight or less and preferably 13 % by weight or less.

30 In addition to the components above-mentioned, there may be blended, as necessary, (i) juice of fruit (concentrated juice of fruit) such as grapefruit, apple, orange, lemon, pineapple, banana, pear, grape or the like, (ii) amino acids (sodium glutamate, glycine, alanine, sodium aspartate and the like), (iii) an inorganic electrolyte serving as a mineral source (sodium chloride, potassium chloride, magnesium chloride, magnesium carbonate, calcium chloride and the like), (iv) vitamins and (v) flavor.

35 Lactosucrose may be used singly as a refined substance as mentioned earlier, but may also be used, in the course of production, as a mixture containing unreacted monosaccharide, disaccharide, oligosaccharide or the like.

The drink composition of the present invention is not particularly limited in intake amount. However, about 0.03 to 0.6 g of lactosucrose per 1 kg of body weight may be generally taken per day.

40 [Industrial Applicability]

The drink composition of the present invention contains lactosucrose as an effective component. Accordingly, the drink composition taken in a human body accelerates an increase in bifid bacteria. This reduces the amount of putrefactive product such as p-cresol, skatole, indole, 4-ethyl phenol or the like to be generated in the intestines. Thus, this is effective in prevention of any of a variety of cancers for which such an intestinal putrefactive product might be a promoter. In the drink composition of the present invention, lactosucrose which is an effective component, is indigestible oligosaccharide and low in calorific value. Accordingly, the drink composition of the present invention may be suitably used as low-caloric food.

50 [Examples]

The following description will discuss the drink composition of the present invention with reference to examples thereof.

55 Test Example

According to the following method, there were measured variations of the amount of a putrefactive product in fecal matter by the administration of lactosucrose of the present invention.

(1) Matter to be Tested

As raw materials, sucrose and lactose were mixed to prepare a mixture, on which β -fructofranosidase was acted. Through respective steps of decoloration, desalinization, filtration and drying, there was prepared a powder preparation 5 containing 59.0 % by weight of lactosucrose (hereinafter referred to as LS55P). The LS55P contained 59.0 % by weight of lactosucrose, 22.7 % by weight of lactose, 8.4 % by weight of sucrose, 1.6 % by weight of fructose, 0.8 % by weight of glucose, 6.8 % by weight of other sugar, and 0.8 % by weight of water.

(2) Persons to be Tested

10 As persons to be tested, there were designated 13 chronically constipated long-stay patients of 55 years and over who had basal diseases such as cerebral infarction, diabetes mellitus and the like. Eleven patients out of these 13 patients had been addicted to the use of a laxative before the test started. During the test period, the dosages of such a laxative were minimized in order to make the physiological operations of these patients highly precise.

15 (3) Intake of the Matter to be Tested

20 The test period extended over four consecutive weeks. The first one week served as a control period during which the matter to be tested was not taken, and the three weeks subsequent to the control period, served as an intake period during which the matter to be tested was taken. The daily dosage of the LS55P was set to 0.32 g/kg B.W. Throughout the intake period, the matter to be tested, to be daily taken was divided into two equal portions, which were taken, as dissolved in about 100 ml of city water, at 10 am and 3 pm, respectively, by each person to be tested.

25 (4) Analysis

30 After each evacuation of each person to be tested, the fecal matter and the urine were separated from each other, and all the amount of fecal matter was collected and measured for weight. After fully kneaded and made homogenous, each fecal matter was preserved under conditions of not greater than -30°C and subjected to analysis of putrefactive product therein. Such analysis was conducted for all fecal matter evacuated during the test period.

35 After the fecal matter and the putrefactive product therein were subjected to variance analysis in a two-way layout, a significant test was conducted on the concentration of the putrefactive product in the fecal matter and the amount of the fecal matter by the Tukey multiple comparison method.

(5) Measuring Method and Results

40 The putrefactive product in the fecal matter, i.e., p-cresol, 4-ethylphenol, indole and skatole were analyzed according to the following method.

45 About 2 g of each fecal matter as precisely measured, was put in a 200-ml Kjeldahl flask, and about 10 ml of purified water was added to the flask, which was then fully suspended. A suitable amount of 2N-sodium hydroxide solution was added to each resulting suspension to adjust pH in the range from 8.5 to 9.0. The solution was then subjected to steam distillation, and about 95 ml of distillate was collected. Purified water was added to this distillate such that the total amount was accurately equal to 100 ml. The distillate was then analyzed for a putrefactive product with the use of gas chromatography mass analyzer (GC-14A gas chromatography interfaced with a GC-MS QP1000EX mass spectrometer manufactured by Shimazu Corporation). The following shows the analyzing conditions:

50 Column: SHIMAZU HiCAP CBPI-M25-025
 Carrier gas: Helium 0.75 kg/cm²
 Inlet port temp.: 250°C
 Column temp.: 50 to 200°C (30°C/min.)
 Ionization method: EI
 Ionization voltage: 70eV
 Separator temp.: 270°C
 Ion source temp.: 250°C

55 After the analysis of the putrefactive product in each fecal matter, there was calculated, for each person to be tested, the total amount of the putrefactive products during the control period, during the first intake week, during the second intake week and during the third intake week. This total amount was regarded as the amount of the putrefactive products evacuated in the fecal matter. For each person to be tested, the concentration of the putrefactive product in fecal matter was calculated from this evacuated amount of putrefactive product and the total weight of the fecal matter

evacuated during the test periods. Table 1 shows variations of the concentration of the putrefactive product in the fecal matter and the amount of the putrefactive product.

The results are shown in terms of the averages \pm standard deviation. In Table 1, significant differences P with respect to the values in the control periods are shown in the following manner. That is, "*" represents that P is smaller than 0.05, "**" represents that P is smaller than 0.01 and "***" represents that P is smaller than 0.001.

In Table 1, the amount of putrefactive product per 1g of fecal matter is shown in the upper row, while the amount of putrefactive product evacuated in the fecal matter per week is shown in the lower row.

Table 1 (1/2)

		Control Period	First Intake Week
p-Cresol	(nmol/gwet)	449.6 \pm 297.0	309.1 \pm 243.8**
	(μ mol/week)	269.8 \pm 148.5	224.7 \pm 113.8
Indole	(nmol/gwet)	125.4 \pm 102.3	91.7 \pm 63.3*
	(μ mol/week)	80.9 \pm 54.2	77.0 \pm 48.7
Skatole	(nmol/gwet)	164.7 \pm 230.1	100.1 \pm 185.4
	(μ mol/week)	94.6 \pm 125.0	59.1 \pm 87.4
4-Ethylphenol	(nmol/gwet)	10.3 \pm 5.5	8.2 \pm 5.2
	(μ mol/week)	6.9 \pm 4.1	6.7 \pm 4.1

Table 1 (2/2)

		Second Intake Week	Third Intake Week
p-Cresol	(nmol/gwet)	277.5 \pm 218.3***	227.1 \pm 179.7***
	(μ mol/week)	205.4 \pm 146.5	175.4 \pm 113.6
Indole	(nmol/gwet)	89.8 \pm 72.0*	83.3 \pm 34.9**
	(μ mol/week)	80.0 \pm 71.7	76.7 \pm 47.7
Skatole	(nmol/gwet)	74.5 \pm 131.0**	66.2 \pm 135.2**
	(μ mol/week)	56.3 \pm 99.1	49.0 \pm 88.9*
4-Ethylphenol	(nmol/gwet)	10.7 \pm 5.9	8.5 \pm 4.7
	(μ mol/week)	8.2 \pm 4.1	7.1 \pm 4.3

Examples 1 to 11 (Drink Composition)

In each of Examples 1 to 11, a healthful drink composition was prepared by mixing the components shown in Table 50 2 and adding water to the resulting mixture such that the amount thereof was equal to 100 ml. In Table 2, each gas volume value refers to an index representing the amount of contained carbon dioxide. Accordingly, as the numeral of gas volume is increased, the amount of contained carbon dioxide is increased. More specifically, the gas volume value is determined such that, when a solution contains gas of carbon dioxide in the same volume as that of the solution, the gas volume value of the solution is equal to 1.

Table 2 (1/2)

Component (in 100 ml)	Example No.					
	1	2	3	4	5	6
Lactosucrose (g)	3	12	9	1	8	2
Glucide (g)						
Isomerized sugar	8	-	-	7	7	8
Sucrose	-	1	8	3	7	5
Fructose	2	6	-	1	-	-
Glucose	2	2	-	-	2	-
Buffer agent (mg)						
Citric acid	3	2	-	-	8	-
Tartaric acid	-	2	-	2	-	-
Malic acid	4	-	8	-	5	-
Lactic acid	8	-	-	2	-	20
Sodium citrate	20	30	10	-	80	-
Sodium tartrate	60	-	-	60	25	70
Sodium malate	-	80	150	-	-	100
Calcium lactate	-	-	-	-	15	10
Inorganic Electrolyte (mg)						
Sodium chloride	-	-	4	-	1	1.5
Potassium chloride	-	3	-	2	-	-
Magnesium chloride	2	-	1	-	-	-
Fruit juice (%)	3	-	1	0.5	0.1	-
Flavor & Sweetner				Suitable quantity		
Gas volume	-	-	-	-	-	3.0
pH	5.0	6.3	5.8	4.9	5.8	5.3

5

10

15

20

25

30

35

40

45

50

55

Table 2 (2/2)

	Component (in 100 ml)	Example No.				
		7	8	9	10	11
5	Lactosucrose (g)	4	10	15	7	13
10	Glucide (g)					
15	Isomerized sugar	5	-	-	9	-
20	Sucrose	3	-	-	-	-
25	Fructose	3	-	-	-	4
30	Glucose	3	-	-	2	-
35	Buffer agent (mg)					
40	Citric acid	-	2	5	-	-
	Tartaric acid	-	-	-	10	-
	Malic acid	4.5	-	-	-	-
	Lactic acid	-	-	-	10	-
	Sodium citrate	-	100	55	70	-
	Sodium tartrate	-	-	-	30	20
	Sodium malate	45	-	10	-	50
	Calcium lactate	-	-	-	-	5
	Inorganic Electrolyte (mg)					
	Sodium chloride	-	-	2	-	-
	Potassium chloride	5	-	1	1	-
	Magnesium chloride	-	-	1	-	-
	Fruit juice (%)	-	-	-	2	0.3
	Flavor & Sweetner			Suitable quantity		
	Gas volume	2.0	2.5	2.3	3.3	1.5
	pH	5.5	5.6	6.4	5.6	5.9

45 Claims

1. A drink composition for inhibiting the formation of an intestinal putrefactive product, containing lactosucrose in an amount of 0.5 to 30 g/100 ml and a buffer solution being added to said composition such that the pH of said composition is adjusted in the range from 4.0 to 6.5.
2. A drink composition according to claim 1, wherein the buffer solution comprises a weak acid having a buffer function and a salt thereof.
3. The use of a drink composition according to claim 1 for preparing a medicament for inhibiting the formation of an intestinal putrefactive product.
4. The use according to claim 3 wherein the buffer solution comprises a weak acid having a buffer function and a salt thereof.

5. A process for preparing a drink composition for inhibiting the formation of an intestinal putrefactive product by mixing at least lactosucrose in an amount of 0.5 to 30 g/100 ml and a buffer agent in an amount such that the pH of said composition is adjusted in the range of 4.0 to 6.5.

5 Patentansprüche

1. Trinkzusammensetzung zur Hemmung der Bildung eines intestinalen Fäulnisproduktes, die Lactosucrose in einer Menge von 0,5 bis 30 g/100 ml und eine Pufferlösung, die der Zusammensetzung zugesetzt ist, so daß der pH der Zusammensetzung auf einen Wert im Bereich zwischen 4,0 und 6,5 eingestellt ist, enthält.
- 10 2. Trinkzusammensetzung nach Anspruch 1, in der die Pufferlösung eine schwache Säure, die eine Pufferwirkung hat, und ein Salz derselben enthält.
- 15 3. Verwendung einer Trinkzusammensetzung nach Anspruch 1 zur Herstellung eines Arzneimittels zur Hemmung der Bildung eines intestinalen Fäulnisproduktes.
4. Verwendung nach Anspruch 3, wobei die Pufferlösung eine schwache Säure, die eine Pufferwirkung hat, und ein Salz derselben enthält.
- 20 5. Verfahren zur Herstellung einer Trinkzusammensetzung zur Hemmung der Bildung eines intestinalen Fäulnisproduktes durch Vermischen von mindestens in einer Menge von 0,5 bis 30 g/100 ml und einem Pufferagens in einer solchen Menge, daß der pH der Zusammensetzung auf einen Wert im Bereich zwischen 4,0 und 6,5 eingestellt ist.

Revendications

1. Composition de boisson pour inhiber la formation d'un produit de putréfaction intestinale, contenant du lacto-saccharose dans une quantité de 0,5 à 30 g/100 ml et une solution tampon étant ajoutée à ladite composition de façon que le pH de ladite composition soit ajusté dans la gamme de 4,0 à 6,5.
- 30 2. Composition de boisson selon la revendication 1, dans laquelle la solution tampon comprend un acide faible ayant une fonction de tampon et un sel de celui-ci.
3. Utilisation d'une composition de boisson selon la revendication 1 pour préparer un médicament afin d'inhiber la formation d'un produit de putréfaction intestinale.
- 35 4. Utilisation selon la revendication 3, dans laquelle la solution tampon comprend un acide faible ayant une fonction de tampon et un sel de celui-ci.
- 40 5. Procédé pour préparer une composition de boisson pour inhiber la formation d'un produit de putréfaction intestinale en mélangeant au moins du lacto-saccharose dans une quantité de 0,5 à 30 g/100 ml et un agent tampon dans une quantité telle que le pH de ladite composition est ajusté dans la gamme de 4,0 à 6,5.

45

50

55